Research Licence Renewal Inspection Report

Project Title	Genetic analysis of human preimplantation embryos
Centre Name	Edinburgh Fertility and Reproductive Endocrine Centre,
Centre Number	0201
Research Licence Number(s)	R0181
Centre Address	Assisted Conception Programme, Edinburgh Fertility and Reproductive Endocrine Centre, Edinburgh Royal Infirmary, Old Dalkeith Road, Edinburgh
Donating treatment Centre numbers	0201
Inspection date	22 April 2008
Licence Committee Date	18th June 2008
Inspector(s)	Andrew Leonard Grace Cunningham
Fee Paid – date (if applicable)	Fee paid
Person(s) Responsible	Dr Susan Pickering
Nominal Licensee	Dr K Joo Thong
Licence expiry date	Renewal Licence Application; current expiry 31/07/08

About the Inspection:
The purpose of the inspection is to ensure that research is carried out in compliance with the HF&E Act 1990, Code of Practice, licence conditions and directions and that progress is made towards achieving the stated aims of the project.

The report is used to summarise the findings of the inspection highlighting areas of firm compliance and good practice, as well as areas where improvement may be required to meet regulatory standards. It is primarily written for the Licence Committee who makes the decision about the Centre's licence renewal application. The report is also available to patients and the public following the Licence Committee meeting.

This report covers the period between 1 August 2007 and 31 March 2008.

Brief Description of the Project

R0181, **Genetic analysis of human preimplantation embryos**
Preimplantation Genetic Diagnosis (PGD) is a technique used to test early human embryos for the presence of a specific genetic disease before implantation into the womb and is useful for couples who have a family history of genetic disease. To carry out PGD, sophisticated genetic analysis of single cells is required and such tests have not always been reliable or consistent and have also proved extremely costly to develop. A new technique has recently been discovered which greatly enhances the amount of genetic information which can be generated from a single cell and use of this technique may simplify the PGD procedure significantly. The aims of this project are to apply this new technology to single cells and then to expand substantially the downstream genetic analysis using high-density genotyping. The reliability and accuracy of such methods on single embryonic cells will be assessed using SNP haplotype tests for aneuploidy (already developed for prenatal use) and for Cystic Fibrosis (in development)

The project is licensed under the following purposes from Schedule 2 of the Human Fertilisation and Embryology Act 1990, 3(2)(e) developing methods for detecting the presence of gene or chromosome abnormalities in embryos before implantation, and from the Human Fertilisation and Embryology Regulations 2001, s2(a) Increasing knowledge about the development of embryos.

Research activities	Research on human embryos	✓
	Storage of licensed material	
	Creation of embryos for research	
	Derivation of human embryonic stem cells	
	Cell nuclear replacement	

Other specific activities involved in the project:
- **i)** Embryo biopsy/embryo disaggregation
- **ii)** Lysis of embryos followed by PCR and genetic analysis of embryos/blastomeres

Summary for Licence Committee

The renewal inspection pertained to Project R0181 at HFEA Centre 0201. Project R0181 investigates the use of multiple displacement amplification (MDA) followed by single nucleotide polymorphism (SNP) marker analysis, as a method for performing Preimplantation Genetic Diagnosis (PGD). The applicants consider that the development of MDA/SNP marker analysis for PGD would greatly enhance the amount of genetic information generated from a single blastomere and may simplify the PGD procedure significantly. The aims of this project are to apply this new technology to single cells and then to expand the downstream genetic analysis using high-density genotyping.

The applicants have considerable appropriate experience and are well qualified to continue the research project. The proposed premises and equipment are appropriate and procedures are in place to ensure that patients are treated respectfully and their consent is not breached. The donation to research procedure complies with the Code of Practice, 7th edition. The research project has the approval of the local research ethics committee. The PR has appropriately completed the PR Entry Programme.

Embryo usage in project R0181 this year was reported in the pre-inspection questionnaire. In summary, the project has used 16 embryos (14 fresh and 2 frozen) since 1st August 2007. The project has progressed in that only 20% of markers were amplified in early experiments, whereas efficiencies between 75 and 100% have recently been obtained. Progress has though been slower than expected. Some time was taken to implement changes requested by Licence Committee when the licence was granted and time for research activity has been limited since two embryology staff left the Centre; these posts have only recently been filled. Because of these issues, the PR considers they have effectively had only 4 months in which to perform research hence this application for a further year's licence, during which the PR expects the project to be completed.

The application for R0181 was considered by an external reviewer who supported renewal of the project research licence. The limited work preformed in the last year was noted and the reasons for this provided by the PR were considered justified.

The inspectorate recommend renewal of the research licence for project R0181 for a period of 1 year.

Report of Inspection findings

1. **Organisation**

Desired Outcome: The research is well-organised and managed and complies with the requirements of the HFE Act.

Summary of findings from inspection
Evidence of:

- Leadership and management
- Staffing
- Funding
- Organisation of the Centre
- Resource management
- Research governance

Staff R0181

Principal investigator	Dr Susan Pickering
Scientists	1 other at Centre 0201, 2 collaborators at Western General Hospital, Edinburgh, are on the licence for R0181
Support staff (receptionists, record managers, quality and risk managers etc)	Staff at Centre 0201

Highlighted areas of firm compliance
The PR has extensive knowledge of the regulatory requirements of the HFEA and is the Laboratory Manager but not the PR under the Treatment and Storage Licence. The PR has a research PhD, and has published research papers. The PR is also an experienced embryologist and has extensive knowledge of preimplantation genetic diagnosis. The Nominal Licensee, is a consultant subspecialist in reproductive medicine and consultant obstetrician and gynaecologist. He has held the post of PR of the treatment and storage licence at Centre 0201 since 1998. The management and co-ordination of resources is shown on the organisational chart within the quality manual. Inspection of the Centre and discussion with staff indicated that the Centre and its laboratory are well organized and all activities are governed by an extensive quality manual. The quality management system is certificated to ISO 9001:2000. The Centre holds weekly all-staff meetings, covering all aspects of the treatment and storage activities as well as the research project. Minutes are circulated to all staff. The Research PR has presented data from the project at these meetings. The PR carries out embryo biopsy for the project, though another embryologist is being trained in biopsy and assists with embryo receipt into the research programme. Once isolated, the blastomeres are taken to a non-embryology laboratory within Centre 0201, where they are placed into tubes for lysis followed by Multiple Displacement Amplification of their DNA. The amplified embryonic DNA is transferred to clinical scientists from Western General Hospital, Edinburgh for genetic analysis. The clinical scientists are HPC registered.

The Centre has been granted a small project grant from NHS Lothian Research and Development fund to purchase consumables and a dedicated PCR machine for the project. The research project utilises the general laboratory facilities at Centre 0201, which are secure and well supplied and maintained and contain all the equipment required to complete their work on the project. The Centre aim to apply for support from NHS Lothian to develop PGD for single gene defects as a clinical service. This would include support for the development of new tests and assays at the Western General Hospital, using amplified embryonic DNA donated and collected at Centre 0201 under this licence.

Research activities are controlled by written procedures, some supplied prior to inspection, which ensure a consistent approach and that patient consent is collected appropriately. The PR and/or research staff present to all staff seminar meetings at the Centre 4 times per year to maintain the project's profile among the staff and to feedback results.

Issues for consideration
None
Executive recommendations for Licence Committee
None
Areas not covered in by this inspection
All covered

2. Premises and equipment

Desired Outcome: The premises and equipment are safe, secure and suitable for their purpose.

Summary of findings from inspection:

- Suitability of premises
- Storage facilities
- Safety of equipment
- Servicing and maintenance of equipment

Highlighted areas of firm compliance
Manipulation of viable embryos is carried out in the laboratories used for the treatment and storage licence at Centre 0201. At this apparently compliant Centre, these laboratories are well equipped and maintained and access is restricted to licensed personnel only. Laboratory premises have all been recently risk assessed and are inspected annually for Health and Safety compliance. Embryos are not stored under this research licence. As soon as embryos stored under the treatment and storage licence are transferred to the research project, they are thawed, lysed and subjected to Multiple Displacement Analysis to produce DNA for later genetic analysis. This amplified embryonic DNA is stored at Centre 0201 before transfer, it being non-licensed, to the Clinical Scientists at the Western General Hospital. Laboratory equipment at the Centre is, including that used in research, on a database which ensures regular servicing and maintenance. It is used according to manufacturer's recommended protocols and is electrically tested annually.
Issues for consideration
None
Executive recommendations for Licence Committee
None
Areas not covered in by this inspection
All covered

3. Donation of material

Desired outcome: Donors are recruited appropriately and any research carried out on their embryos is in accordance with their consent.

Summary of findings from inspection:

- Recruitment of donors
- Ensuring prospective donors have access to further guidance
- Ensuring prospective donors have time to consider donation properly
- Ensuring patient consent is not breached
- Donor and patient records
- Prevention of coercion of prospective donors

Highlighted areas of firm compliance
Donors of both fresh and frozen embryos are recruited from Centre 0201 only. The recruitment of donors is covered by written procedures which verify documentation and consents are appropriate for research donation and that written information is provided. Donated fresh embryos have abnormally fertilised (0, 1 or 3 PN) or are classified as being of poor quality (Grade 3 or less) on Day 2, when the embryologists select which embryos are suitable for transfer and for freezing. The embryologists involved in research have no role in the embryo grading and selection for transfer or research in research consented cases. Hence research stopping for some of the last year when lab staff numbers were depleted by the departure of two embryologists, priority obviously being given to the remaining staff's clinical work. Patients undergoing IVF attend the Centre for an initial consultation with a doctor at which they are given a general research information sheet on which they sign a statement of interest. Patients have a later meeting with a nurse to discuss treatment, at which research projects carried out at the Centre are discussed with those who have signed the statement of interest. If the couple remain interested, they are given written research information to inform their decision and a consent form. Contact details are provided in the patient information for a designated staff member as the contact point for further information. The consent forms are completed and returned when the patient attends the Centre for their pre-treatment scan, when the consents are checked by the nurse and placed in the patient's medical notes. Patients then have IVF treatment as normal. Following fertilisation and embryo transfer at Day 2, couples with abnormally fertilised or poor quality surplus embryos suitable for the research project are identified. Two embryologists check the patient's notes to determine whether HFEA and Centre research consents are present. If consents are present, the embryos are handed over to the two research project associated staff in the embryology laboratory. The handover is witnessed on the research embryo log sheet but not in the patient notes or on the laboratory witnessing sheet (see below). Donated material is marked with an effective anonymised code by the Centre 0201 research staff. All embryos donated are also entered into a research project embryo log which details the patient Centre number, biopsy procedure, lysis and a second anonymised code allocated by the genetic researchers at Western General Hospital, Edinburgh on collection of amplifed blastomere DNA. Patients with embryos in storage are sent an annual letter asking whether they wish to continue storage for another year and outlining the options available. If a couple state that they wish to donate their embryos to research, they are asked to attend the clinic to discuss the research project. If, after this discussion, they still wish to donate embryos to research, the

couple are provided a detailed research information sheet and consent forms. Couples are given the opportunity to take these forms home to read and discuss amongst themselves. The couple then complete the research consent forms which are checked by two embryologists. The research consent does not rescind the storage consent and the Centre sometime leave the embryos in storage until they can be processed. At this point the embryos are removed from the storage dewars, thawed and handed over to the researchers. This procedure is witnessed on the research embryo log sheet but not in the patient notes or on the laboratory witnessing sheet. The thawing is however noted on the patient laboratory record sheet.

Donated material is marked with an anonymised code by the Centre 0201 research staff and allocated an embryo log sheet. A different anonymising code is then allocated by the genetic analysts from Western General Hospital. Both codes are logged with other information related to the donation and the biopsy in the project R0181 embryo log at Centre 0201. This log is kept securely in the PR's office and allows tracking from research records to patient records.

Given the practices described by the PR, donors were considered by the inspectorate to be provided enough time to consider their donation. No complaints regarding this, or coercion, have been received by the HFEA or the Centre. Donors receive no benefits.

Issues for consideration

Research and clinical embryology are performed in the same laboratory. The PR should write a procedure in case of her absence and for new staff induction and use, which reflects the Centre's working practices, to ensure compliance with Code of Practice, 7th edition, Standards, S.8.4.1. While their working practices achieve the required separation of research donation from clinical embryology, these practices must be documented. This separation should also be considered in a review of the procedure for disposal of embryos which are coming to the end of their consented storage period. This was a requirement of the initial research licence inspection, endorsed be a Licence Committee. It had not been done because the PR considered the working practices which ensured separation were sufficient.

The Centre should ensure that the transfer of embryos to research is witnessed in the patient notes as well as in the research records, so that the patient notes provide a full account of the patients treatment and the processes that their gametes and embryos have passed through.

Executive recommendations for Licence Committee

That the PR should write a procedure in case of her absence and for new staff induction and use, which reflects the Centre's working practices regarding maintaining an effective separation between research and clinical embryology, to ensure compliance with Code of Practice, 7th edition, Standards, S.8.4.1.

That the PR ensure that the witnessing of embryo transfer from treatment to research is documented in the patient notes to ensure compliance with witnessing guidelines (Code of Practice, 7th edition, Guidance, G.13.1.1 and Licence Condition A.2.5 on the Centre's Treatment and Storage licence.

Areas not covered in by this inspection

All covered

4. Patient information and consents

Desired outcome: Patients are provided with appropriate information which allows them to give informed consent.

Summary of findings from inspection:

- Patient information
- Consent forms
- Patient information for projects deriving embryonic stem cells
- Consent forms for projects deriving embryonic stem cells

Outcome of audit of records
An audit of 4 patient records showed that all research consents were collected appropriately, with one exception (discussed below), and the transfers to research had been witnessed appropriately in all cases. In addition, the embryos transferred to research were seen to be of a quality which complied with the selection criteria outlined in the donation procedure and in the patient research information sheet.
Summary
Patient information and consent forms for project R0181 were reviewed by the inspectorate. They were compliant with the Code of Practice, 7th edition, and were considered well prepared and informative, except for the specific issues raised below. There is no stem cell derivation associated with donation to this research project.
Issues for consideration
During the notes audit, in one set of notes it was seen that dates of signatures indicated that research interest and then full consent had been obtained in a process which had started only 3 days before oocyte collection. This was contrary to the Centre's procedures for donor recruitment, as verbally described by the PR. Patient information should provide details for a person through whom consent can be withdrawn or varied. It instead instructs patients to speak to anybody on the Centre staff. Contact details are provided in the patient information, for an individual who can provide further information about the research project but it is not discussed that this is a contact for withdrawing or varying consent. At the request of the local research ethics committee, patient information contains the statement: *"if requested, results of this research can be provided for your information."* The inclusion of this statement and its meaning has been previously discussed with the Centre at the initial project inspection. The PR clarified that the only information fed back to patients is general information related to the outcome of the research project. Patients are not given specific information regarding the analysis of their embryos. This is verbally clarified with the patients by the nurses when the research project is discussed with them.
Executive recommendations for Licence Committee
That the Centre review the research donor recruitment policies and procedure and ensure they are compliant with the Code of Practice, 7th edition, and that the Centre in their working practices adhere to their procedures. Specifically, patients must be given enough time to

consider information provided to them such that consent is fully informed and consideration of it is not rushed. In normal practice this general means that in normal practice, patient information should be provided in the early phases of the treatment cycle in which donation is to be made, or earlier.

That the Centre review the patient information and consent forms for project R0181 given the issues outlined above; specifically patient information and consent forms describe the provision for patients to withdraw or vary their consent, but they should also provide contact details for a named individual through whom this can be achieved, to be compliant with Code of Practice, 7th edition, G.5.13.1 (g).

Areas not covered in this inspection

None.

5. Scientific practice R0181 - Genetic analysis of human preimplantation embryos

Desired outcome: Research is carried out in accordance with licence conditions and makes progress towards achieving stated aims

Summary of:

- Peer review

Summary
The PR stated in the pre-inspection questionnaire that project R0181 had utilised 16 embryos between 01/08/2007 and 31/03/2008. In summary, the project has progressed in that only 20% of markers were amplified in early experiments, whereas efficiencies between 75 and 100% have recently been obtained. Progress has been slower than expected. Some time was taken to implement some minor changes requested by Licence Committee (listed in the attached Licence Committee minutes in Appendix B) when the licence was granted. In addition, time for research has been limited since two embryology staff left the Centre; these posts have only recently been filled. Effectively the PR considers only 4 months work has been accomplished, hence this application for a further year's licence, during which the PR expects the project to be completed. The research team predict that the project will use 30 fresh embryos and 15 frozen embryos next year.
Summary of audit of stored and biopsied material
No embryos are stored under this project licence
Renewed project objectives
Future work The PR states in the renewal application: 'As we have only carried out 4 months work on this project, we have not yet carried out all the work which we detailed in our original application. We are hoping to be able to fulfil our original objectives within the original timescale, however, we are applying for a renewal for a further year due to the considerable delay we have suffered due to shortage of staff in the assisted conception unit. It is expected that once we have completed the work in the original application, we are unlikely to require another research licence.' The application also pointed out that no new work is planned which is not included in the original application and that the project objectives have remained the same as on original application, i.e.: Objective 1: To assess the reliability of SNP haplotypes at any genetic location and compare the results obtained with those from more conventional genetic assays. Objective 2: To design locus specific SNP haplotyping assays for a key disease gene, Cystic fibrosis transmembrane conductance regulator (CFTR).

Objective 3:
To examine the chromosome constitution of abnormally fertilised embryos

Summary of research undertaken

As supplied by the Licence Renewal application:

The objectives of the initial application were as follows:

- To assess the reliability of SNP haplotypes at any genetic location and compare the results obtained with those from more conventional genetic assays.
- To design locus specific SNP haplotyping assays for a key disease gene, Cystic fibrosis transmembrane conductance regulator (CFTR).
- To examine the chromosome constitution of abnormally fertilised embryos

With regard to the first objective we have successfully amplified DNA from several embryos using MDA PCR and subsequently analysed this DNA using two complex downstream genetic assays. These are discussed in detail below. We consider this to be the first step towards our eventual aim, which is to carry out highly sophisticated SNP haplotyping assays, using several different real time PCR platforms available to us in Edinburgh. This objective has not yet been attempted, but using some of the PCR products already in our possession, we hope to be starting this work within the next three months, still well within the period of our initial licence application.

With regard to the second objective, we have developed a new specific CF assay which will be used for clinical PGD in the future. We now need to compare results obtained with this assay with those obtained with SNP haplotyping assays. It is hoped this will be carried out in the next 6 months

With regard to the third objective, we have so far only been able to obtain two abnormally fertilised embryos for analysis, but are hoping to target this group of embryos more specifically in the next six months.

i) *Research undertaken to date.*

16 embryos have been used in this project, 14 fresh embryos donated by patients undergoing assisted conception cycles and 2 from a patient who donated their frozen embryos (no longer require for clinical use). Three embryos have been collected intact and the rest have been biopsied and one or two cells removed for genetic analysis. The remainder of the embryo (without zona pellucida) has then been collected as a single sample and analysed for comparison with the genetic fingerprint obtained from the biopsied cell(s). All samples have been lysed and then amplified using multiple displacement amplification (MDA) PCR according to manufacturers instructions, published protocols and advice from the Guy's hospital PGD team.

The amplified DNA from each sample has been subjected to further diagnostic tests, during which subsequent PCR reactions have been carried out using specific primers to amplify several different loci within the genome. Two different genetic assays have been applied to each sample. One assay is currently used routinely in the Genetics laboratory at the Western General for the diagnosis of aneuploidy in prenatal samples. This consists of 12 markers, four markers each on chromosomes 13, 18 and 21. Details of the specific markers and primers used to amplify each sequence can be provided on request. Several markers in this assay were originally used by Mann et al for the rapid prenatal diagnosis of aneuploidy using quantitative fluorescence-PCR (Mann et al 2005). Others have been developed in house and used for many years for routine diagnostic work. The second assay is for the detection of mutations and linkage within the cystic fibrosis (CF) gene. The assay consists of 11 markers used originally by the PGD team at Guy's hospital for diagnosis of CF using MDA PCR followed by genetic haplotyping (Renwick et al 2006). In addition, 4 new markers which are likely to be more informative in the Scottish population have been developed in house as part of this research project, with a view to carrying out clinical PGD for CF in the future.

For each sample, the amplification efficiency and allele drop out rate (where it can be detected) at each genetic locus has been determined. The allele sizes, peak heights and characteristics have also been compared for each locus where amplification was successful and signal from biopsy sample and embryo compared.

ii) *Results*

Following successful amplification of test samples over a period of weeks (buccal cells and diluted genomic DNA samples), initially, three embryos were collected intact (minus zona pellucida) and two embryos biopsied and subjected to diagnosis using both aneuploidy and CF assays as a test of the conditions on embryonic material. Although conditions used were identical to those previously used and those used in published protocols and recommended in the manufacturers instructions, relatively little PCR amplification was observed at any locus. Amplification was seen at only 19 loci from a possible 96 (20%), which given that 60 of these tests were carried out on MDA PCR DNA originating from multiple cell samples (intact embryos or the remains of a biopsied embryo) was very poor compared to previous results with single buccal cells and other published results (Renwick et al 2006, 2007; Hellani at el 2004; Lledo et al 2006; Renn et al 2007). Following tests of various reagents on buccal cells another three embryos were biopsied and subjected to analysis, this time using only the aneuploidy assay. Again, disappointing results were obtained, with a relatively low amplification efficiency and very high allele drop out rate observed, results which are not appropriate for an accurate and reliable clinical PGD assay.

Eventually, following several more reagent tests, discussions with manufacturers and the Guy's PGD team, it was discovered that the MDA phi 29 polymerase performs more reliably if stored at –80^{o}C, despite the manufacturer recommending storage at –20^{o}C. A new MDA kit was obtained and two frozen-thawed embryos were biopsied and the samples subjected to MDA-PCR and then both the downstream genetic tests. Results from these tests are detailed in Table 1. The amplification efficiency of individual loci within the CF assay was still low (13% and 47% of loci were amplified in multiple cell samples and 13%, 13% and 7% for single cells)

and allele drop out rate could not be calculated because alleles were not co-incident between samples. However, for the aneuploidy assay, amplification efficiency of individual loci was substantially improved (multiple samples 42% and 92%; single cells 42%, 50% and one with no amplification) and ADO rates were at an acceptable level (0% and 17%). On the basis of these results, a few technical changes were made to both downstream assays (concentration of primers, number of PCR cycles etc) and further improvements were obtained. However, the amplification efficiency and ADO rates were still not deemed good enough for a clinical PGD assay. As these results had been obtained on frozen thawed embryos, which had arrested in their development and therefore may have been of sub-optimal quality, it was decided to subject more fresh embryos to the improved assays and compare the results.

A further 6 embryos were biopsied and collected for MDA-PCR as before and subjected to both downstream genetic assays. The results this time were significantly improved for both assays, although for single cells, on average a higher amplification efficiency and lower ADO rate was seen at loci amplified using with the aneuploidy assay.

For the aneuploidy assay, in four of the multiple samples 100% of loci were successfully amplified and in the remaining two multiple samples, amplification efficiencies of 83% and 92% were obtained. For the single cell samples, amplification efficiencies between 75 and 100% were obtained in five samples, with one sample showing no signs of amplification at any locus. For the CF assay, in five of the multiple samples 100% of loci were successfully amplified and in the remaining sample an amplification efficiency of 87% was obtained. For the single cell samples, amplification efficiencies between 40 and 100% were obtained in five samples, with one sample showing no signs of amplification at any locus. This was the same sample in which there was no amplification using the aneuploidy assay, suggesting complete failure of the MDA-PCR or failure of the operator to successfully place the cell in the sample tube.

Allele drop out rates were more difficult to calculate with certainty, as the parental genotype was unknown, but ADO rates of between 0 and 40% were obtained with the aneuploidy assay and ADO rates of between 13 and 57% were obtained using the CF assay. In general, ADO was lower in the aneuploidy assay. A relatively high level of ADO is generally acceptable when carrying out PGD diagnosis by haplotyping compared to diagnosis by more conventional mutation detection. Once a consistent picture of ADO with any particular assay has been obtained, the number of informative markers which would be required to give >95% confidence of detecting a particular genotype can be calculated (Renwick et al, 2007). Contamination with extraneous DNA was not identified in any sample.

The reason for differences in amplification efficiency and allele drop out rates seen between the two assays is unknown, although it is likely to be due to downstream technical issues associated with each of individual assays. The aneuploidy assay has been in clinical diagnostic use in the WGH lab' for many years where it is used to test amniotic fluid samples, often containing small quantities of DNA. The downstream amplification conditions have therefore been super-optimised over time. The CF assay is new and is likely to require further downstream optimisation before it can be applied clinically. This optimisation work is ongoing

using the MDA products already generated.

Table 1: PCR results from amplification of single biopsied blastomeres and remains of the embryo from which the blastomere was extracted

	CF assay		Aneuploidy assay		
Sample	Proportion of loci successfully amplified	ADO rate	Proportion of loci successfully amplified	ADO rate	
Embryo 1 (4 cells)* Frozen-thawed	2/15 (13%)		5/12 (42%)		
1 cell biopsy from E1	2/15 (13%)	alleles not co-incident	5/12 (42%)	0/5 (0%)	
1 cell biopsy from E1	2/15 (13%)	alleles not co-incident	No amp	N/A	
Embryo 2 (7 cells)* Frozen-thawed	7/15 (47%)		11/12 (92%)		
1 cell biopsy from E2	1/15 (7%)	alleles not co-incident	6/12 (50%)	1/6 (17%)	
Embryo 3 (5 cells)* Fresh	15/15 (100%)		12/12 (100%)		
1 cell biopsy from E3	7/15 (47%)	4/7 (57%)	10/12 (83%)	2/10 (20%)	
Embryo 4 (7 cells)* Fresh	15/15 (100%)		12/12 (100%)		
1 cell biopsy from E4	6/15 (40%)	2/6 (33%)	11/12 (92%)	4/11 (36%)	
Embryo 5 (8 cells)* Fresh	15/15 (100%)		10/12 (83%)		
1 cell biopsy from E5	6/15 (40%)	1/6 (17%)	9/12 (75%)	0/9 (0%)	
Embryo 6 (5 cells)* Fresh	15/15 (100%)		12/12 (100%)		
1 cell biopsy from E6	No amp	N/A	No amp	N/A	
Embryo 7 (5 cells)* Fresh	15/15 (100%)		12/12 (100%)		
1 cell biopsy from E7	12/15 (80%)	5/12 (42%)	11/12 (92%)	2/11 (18%)	
Embryo 8 (3 cells)* Fresh	13/15 (87%)		11/12 (92%)		
1 cell biopsy from E8	14/15 (93%)	2/15 (13%)	12/12 (100%)	4/10 (40%)	

*The number of cells in brackets were collected as a single sample for analysis. For example, embryo 2 was originally a 6-cell embryo and 2 individual cells were biopsied from it and collected as two separate samples. The remaining 4 cells in the embryo were collected together as another separate sample for confirmation of diagnosis.

Peer review comments (if applicable)
The peer reviewer supported this renewal application without reservation
Issues for consideration
NONE
Executive recommendations for Licence Committee
NONE
Areas not covered on this inspection
NONE

Report compiled by:

Name Andrew Leonard

Designation HFEA inspector

Date 9th June 2008

Appendix A: Centre Staff interviewed

PR
1 embryologist

Appendix B: Licence history

Licence for project R0181 was approved by a Licence Committee on 25th July 2007 after there initial application and inspection:

Research Licence Committee Meeting

25 July 2007
21 Bloomsbury Street London WC1B 3HF

MINUTES Item 1

Research Project R0181: Genetic analysis of human preimplantation embryos
Based at Edinburgh Fertility and Reproductive Endocrine Centre (0201)
Initial Research Licence Application

Members:	In Attendance:
Richard Harries – Chair, Lay Member	Marion Witton – Head of Inspection
Clare Brown, Lay Member	Frances Clift, Legal Adviser
Maybeth Jamieson, Consultant Embryologist, Glasgow Royal Infirmary	Joanne McAlpine, Acting Committee Secretary
William Ledger – Professor of Obstetrics and Gynaecology, University of Sheffield	Barbara Lewis, Observer
Rebekah Dundas – Lay Member	

Conflicts of Interest: Maybeth Jamieson declared a conflict of interest with this item and therefore contributed to the discussion but did not take part in any decision making.

The following papers were considered by the Committee:

- papers for Licence Committee (94 pages)
- no papers were tabled.

1. The papers for this item were presented by Debra Bloor, HFEA Inspector. Dr Bloor informed the Committee that this is an initial application for a licence to carry out research into the application of a novel technique for use in preimplantation genetic diagnosis.

2. Dr Bloor informed the Committee that the PR for the research project, Dr Sue Pickering, has considerable research experience.

3. Dr Bloor confirmed that at the time of the inspection the PR Entry Programme had not been completed but it has since been received and assessed as satisfactory.

4. Dr Bloor explained that the application had been considered by two peer reviewers. One peer reviewer agreed that the research would be useful for developing methods for detecting abnormalities in embryos before implantation and increasing knowledge about the development of embryos. The second peer reviewer thought that the small number of embryos studied would not significantly add to the knowledge of embryos but supported the claim that the research would be useful for developing methods for detecting early abnormalities in embryos.

5. Dr Bloor informed the Committee that the research had been approved by the local research ethics committee. During the inspection visit the premises, equipment and laboratory were found to be suitable.

6. Dr Bloor stated that the patient information regarding the research project was comprehensive and accessible but there was a statement included in the patient information which lacked clarity. This statement was discussed at length in the course of the inspection and subsequent to the review of the draft inspection report the PR reported that the statement has now been amended.

7. In response to a question raised by the Committee regarding the number of embryos identified for use in this project, Dr Bloor explained that the figure of 100 embryos was based on what was practically available and not what was required for this study. Dr Bloor confirmed that the centre has acknowledged that the number of embryos needed cannot be predicted at this stage because it is using a novel technique.

8. The Committee questioned whether embryo biopsy was part of the project and who the researcher would be. Dr Bloor confirmed that the PR would undertake all the biopsy work.

The Committee discussed a few suggestions that they felt would be useful to the project, and asked Ms Hopper to communicate the advised suggestions directly with the PR of the project.

- the centre puts documented procedures in place to demonstrate that the selection of embryos for treatment/freezing will not been influenced by research
- the results of the research are monitored and the research study stops if the objectives of the research are achieved prior to the use of 100 embryos, and that HFEA should be informed when the research is concluded successfully

9. The Committee applied the statutory tests in considering the application. To start, the Committee identified the activity under consideration as the use of embryos in research. The Committee noted that this activity is not prohibited under the Human Fertilisation and Embryology Act 1990.

10. The Committee agreed that this activity appears to be necessary or desirable for the following specified purpose:

- developing methods for detecting the presence of gene or chromosome abnormalities in embryos before implantation
  Human Fertilisation and Embryology Act 1990 Schedule 3(2)(e)

- Increasing knowledge about the development of embryos
  Human Fertilisation and Embryology Act 1990 Schedule s2(a)

11. The Committee agreed that they were satisfied that the proposed research could not be undertaken without the use of human embryos.

12. The Committee agreed that they were satisfied with the patient information and consent forms submitted by the centre subject to some minor amendments to the consents form. The Committee advised that to avoid misinterpretation of the following statement: "If requested, results of this research can be provided for your information." The Centre should consider including in the verbal discussions with patients an explanation that they may receive general information about the project but they will not receive specific information about the outcome of any genetic tests carried out on their embryos.

13. The Committee were satisfied that the requirements for granting a licence under section 16 of the Human Fertilisation and Embryology Act 1990 were fulfilled and decided to grant a licence for the research for a period of twelve months.

Signed…….. … Date……..
Richard Harries (Chair)

Appendix C:
RESPONSE OF PERSON RESPONSIBLE TO INSPECTION REPORT

Centre Number 0201

Name of PR Dr Sue Pickering

Date of Inspection 22 April 2008

Date of Response 10th June 2008

Please state any comments regarding the inspection and actions you have taken or are planning to take following the inspection with time scales

I found the comments of the inspectors very useful and also very much valued the discussions we had on the day. Centre 0201 has not previously held a research licence and although the centre has collaborated with other centres and provided research material under other licences, most of the staff have not been involved in research work before. Therefore, as PR, I have had to set up all the systems and protocols currently in place from scratch and although I am myself experienced and have held two research licenses previously while at Guy's Hospital London, I found it extremely useful to have the system I have set up here "interrogated" by two such experienced inspectors and feel it is much improved by the actions I have taken since the inspection and the changes I have made as a result of our discussions. The changes I have made are outlined below:

- I have clarified the role of each embryologist with regard to their daily duties in the "Technical guidelines" protocol and this is reflected in a more detailed "Embryology staff rota" form. These two documents now detail exactly which embryologist is responsible for grading embryos and deciding on their fate with regard to clinical use.
- I have clarified the procedure to be used when transferring material to a research licence in the "Disposal of fresh/frozen embryos" protocol and in the newly written "Genetic analysis of human embryos" research protocol. In both these protocols the separation of the role of the clinical embryologist and the research scientist is clearly defined.
- I have included an additional witnessing check box on the HFEA clinical witnessing form (in addition to the one already present on the research form) so there is traceability from the medical notes to the laboratory records (which are kept separately) to research records regarding transfer of embryos to a research licence.
- I have clarified the criteria used to determine which embryos are suitable for freezing and which are not in the "Embryo cryopreservation" protocol and I have also clarified the definition of "good quality embryo" (ie embryos which are most suitable for clinical use and freezing) in the "Embryo transfer" protocol. I have also defined the scientific basis for such criteria.
- I have added a line regarding withdrawal of consent into the Patient information sheet, naming a specific person they can contact if they wish to withdraw consent. This is the same person who is the named contact for more information on the project, if the patient wishes it. I have also left in the line that states the patient may contact anyone at the centre to withdraw their consent as the patient may have feel there are specific

people in the centre to whom they would prefer to speak, on the basis of their previous interactions. I don't wish to restrict the patient in any way if they really do want to withdraw their consent. I hope you feel this is appropriate.

All these documents have been sent to my inspector, Mr Andrew Leonard. I hope this covers the issues that were discussed at the inspection. However, if there are outstanding issues which I have not covered, I would be very happy to address them.

Susan J. Pickering

2. Correction of factual inaccuracies

Please let us know of any factual corrections that you believe need to be made (NB we will make any alterations to the report where there are factual inaccuracies. Any other comments about the inspection report will be appended to the report).

I believe the report to be an accurate reflection of the inspection and the items discussed by the persons involved. I have found no factual inaccuracies.

Research Licence Committee Meeting

18 June 2008
21 Bloomsbury Street London WC1B 3HF

MINUTES Item 6

Research Project R0181: Genetic analysis of human preimplantation embryos
Based at Edinburgh Fertility and Reproductive Endocrine Centre (0201)
Licence Renewal

Members:
Emily Jackson – Chair, Lay Member
Richard Harries, Lay Member
Maybeth Jamieson, Consultant Embryologist, Glasgow Royal Infirmary
Neva Haites, Professor of Medical Genetics, University of Aberdeen

In Attendance:
Chris O'Toole, Head of Research Regulation
Claudia Lally, Committee Secretary

Providing Legal Advice:
Mary Timms, Field Fisher Waterhouse

Conflicts of Interest: Maybeth Jamieson declared a conflict of interest with this item and left the room . Other members of the Committee declared that they had no conflicts of interest in relation to this item.

The following tabled papers were considered by the Committee:

- papers for the Committee (57 pages)
- no papers were tabled.

1. The papers for this item were presented by Andrew Leonard, HFEA Inspector. Dr Leonard informed the Committee that this project investigates the use of multiple displacement amplification (MDA) followed by single nucleotide polymorphism (SNP) marker analysis, as a method for performing preimplantation genetic diagnosis. It is considered that the development of this PGD technique would greatly enhance the amount of genetic information generated from a single blastomere and may simplify the PGD procedure.

2. Dr Leonard informed the Committee that the Person Responsible for the project has completed a Person Responsible Entry Programme (PREP) assessment to the satisfaction of the Executive. Progress with the project has recently been slow due to staffing issues, with a total of 4 months work being carried out to date.

3. Dr Leonard discussed the findings of the inspection visit. The most significant finding was the discovery that the centre had not always followed its own protocols in relation to when consent is taken from donors to the research. Specifically, evidence was found of one donor who was approached only three days before her egg collection was due. The centre has however responded fully to this issue and has put in place procedures to ensure that it is not repeated. Dr Leonard affirmed that the centre has responded fully to all the findings of the report and has submitted the information required of them.

The Committee's Decision

4. The Committee identified the activity under consideration as the use of donated embryos in research. The Committee agreed that this activity is not prohibited under the Human Fertilisation and Embryology Act 1990.

5. The Committee considered whether the proposed activity appears either necessary or desirable for one or more of the purposes as set out in paragraph 3(2) of Schedule 2 to the 1990 Act or in paragraph 2(2) of the Human Fertilisation and Embryology (Research Purposes) Regulations 2001. The Committee considered the stated aims of the project and the evaluation of the project by the peer reviewer and agreed that in the context of the project of research these activities appear to be necessary or desirable for the following purpose:

- Human Fertilisation and Embryology Act 1990 Schedule 2 paragraph 3(2)(e) to develop methods for detecting the presence of gene or chromosome abnormalities in embryos before implantation

In reaching this decision the Committee took into account the aim of the research to amplify genetic information from single cells to improve techniques for preimplantation genetic diagnosis.

6. The Committee agreed that this use of embryos is necessary for the purpose of the research since the techniques to be developed will be specific to human embryos.

7. The Committee agreed that they were satisfied about the consent forms and patient information.

8. The Committee agreed that the requirements for the grant of a licence under Section 16 of the Human Fertilisation and Embryology Act 1990 were satisfied, and decided to renew the licence for a period of one year, in accordance with the wishes of the centre.

Signed…… … Date……..
Emily Jackson (Chair)